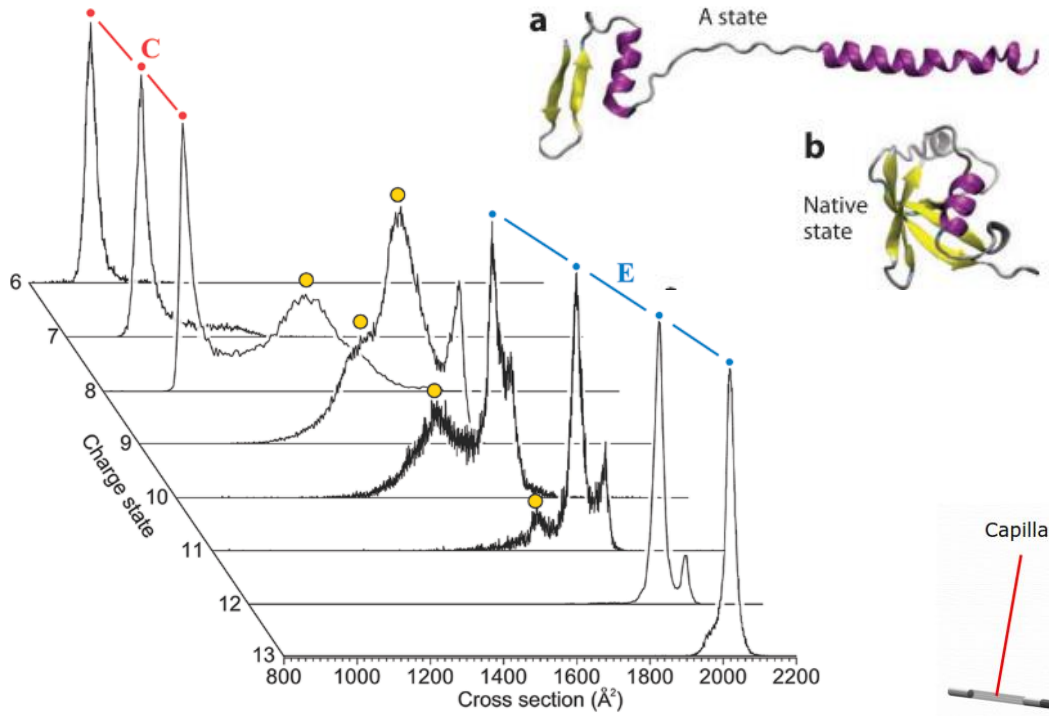
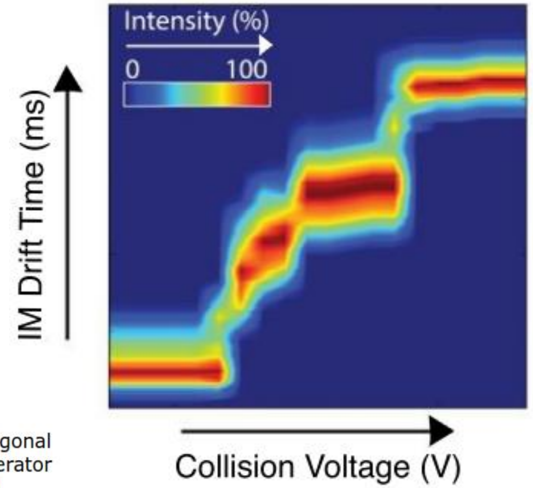
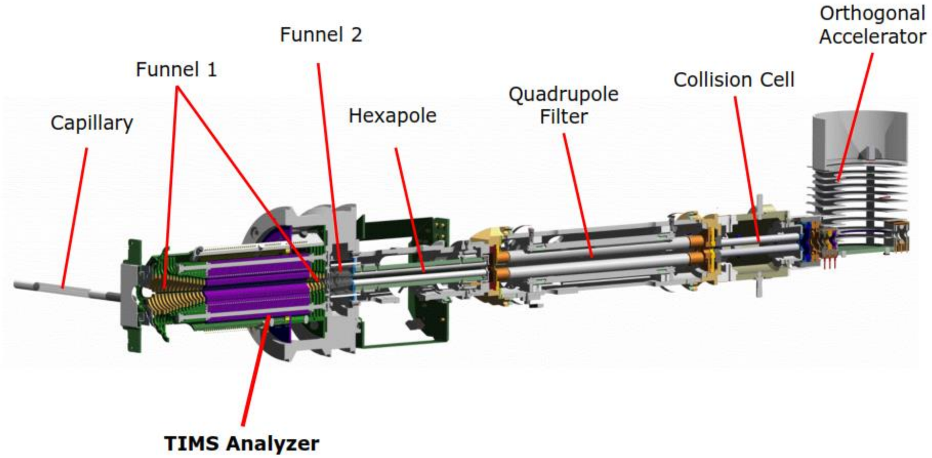
# Advances in mass spectrometry

## Lecture 4 : Ion mobility

Quentin Duez



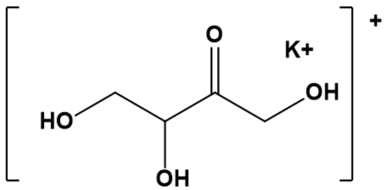
# Outlook

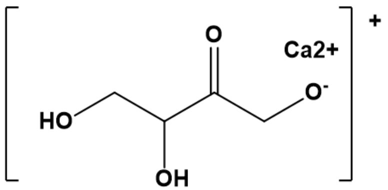
- Why would you do **ion mobility** ?
- Ion mobility : **Principle** of the experiment & **collision cross sections**
- Instrumentation : **DTIMS, TWIMS, TIMS**

# Why would you do ion mobility ?

- Mass spectrometry is sometimes enough …

**Isobaric ions**

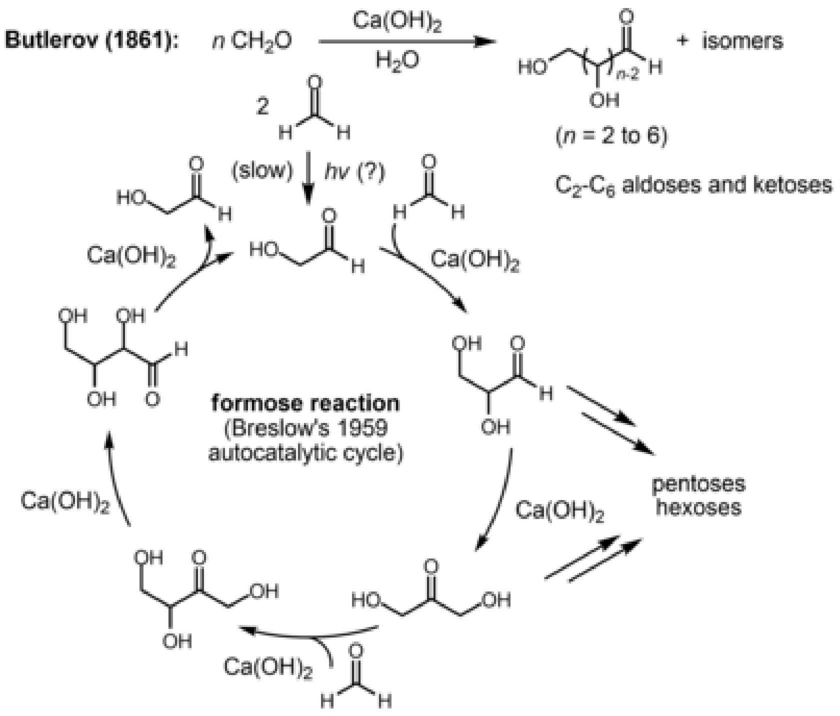


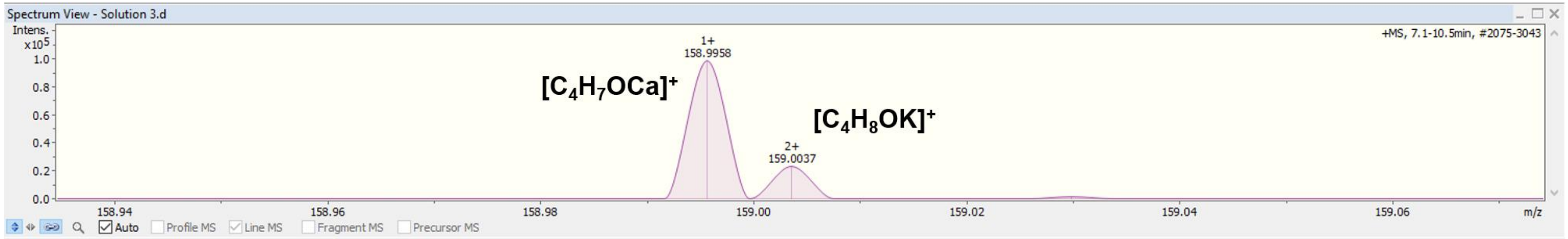




**Mass (H) = 1.008 Da**

**Mass (K) = 38.964 Da**

**Mass (Ca) = 39.963 Da**



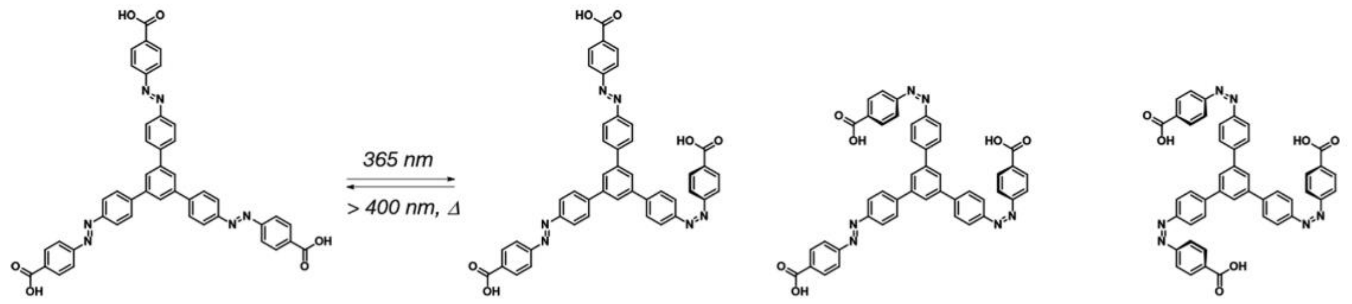




Which mass analyzer was used for this experiment ? Quadrupole or ToF ?

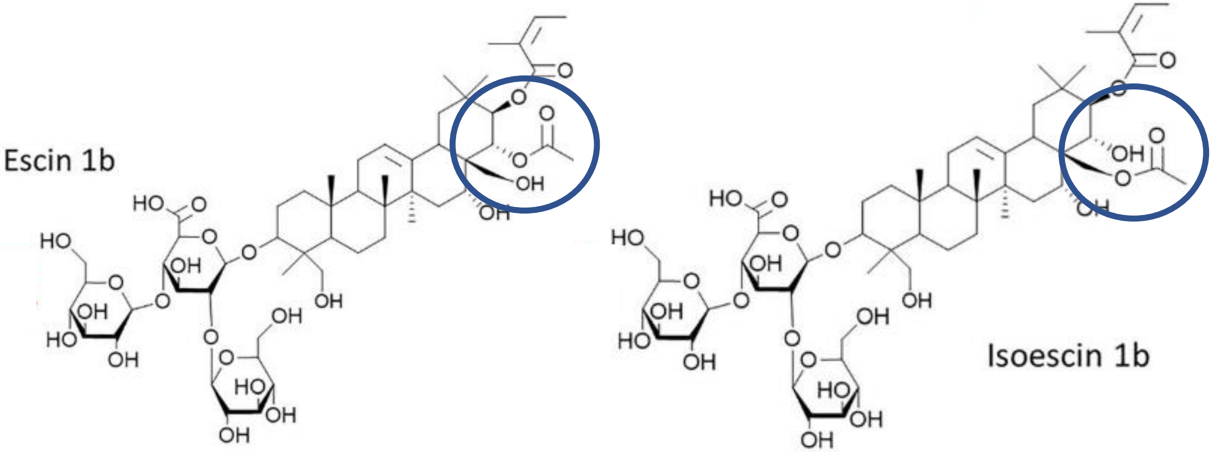
# Why would you do ion mobility ?

- And sometimes not enough …

**Isomeric ions**

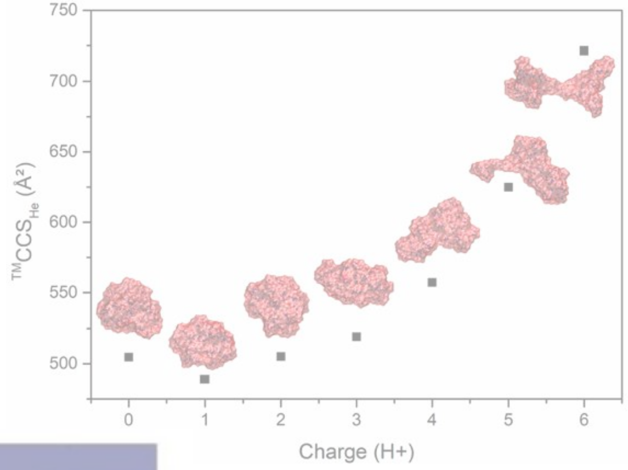


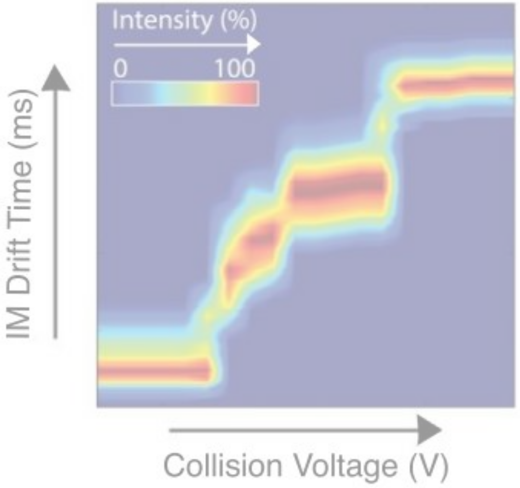

(E,E,E)-1 (E,E,Z)-1 (E,Z,Z)-1 (Z,Z,Z)-1




**Shape of ions in gas-phase**

« PAMAM » dendrimer




Collision-induced unfolding of proteins

Galanti *et al.* (2018) *J. Am. Chem. Soc.*, 140 (47), 16062-16070.
Dixit *et al.* (2018) *Curr. Opin. Chem. Bio*, 42, 93-100.
Colson *et al.* (2019) *J. Am. Soc. Mass. Spec.*, 30 (11), 2228-2237.
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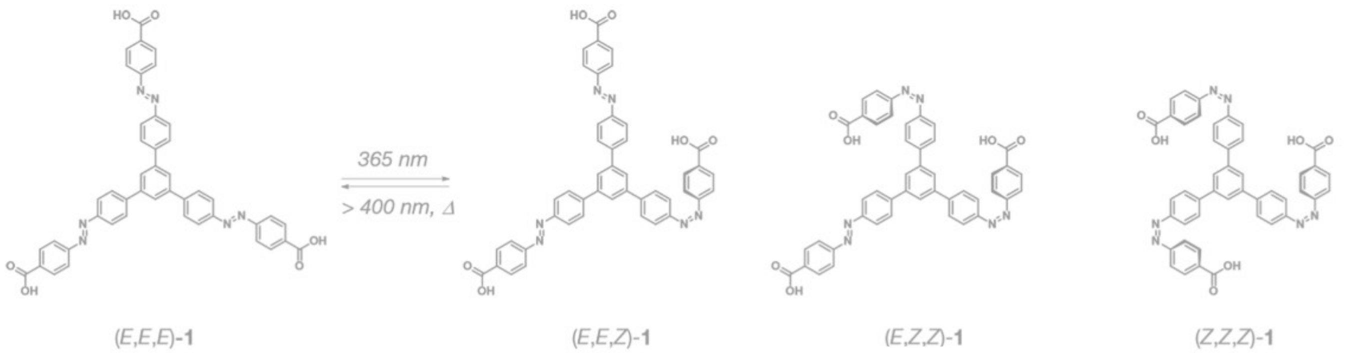
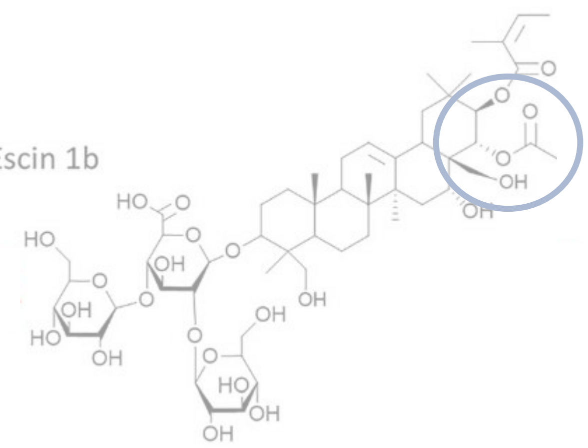
# Why would you do ion mobility ?

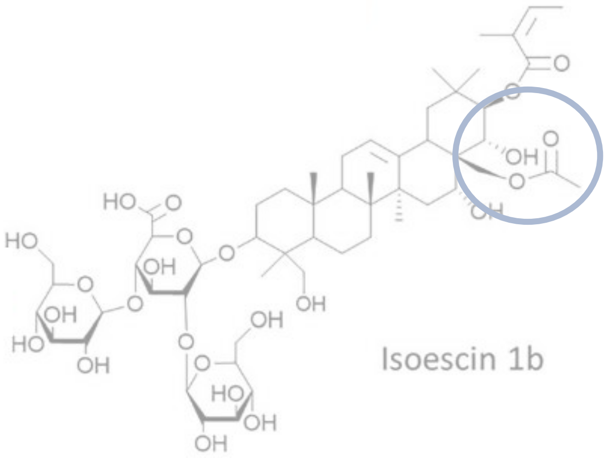
- And sometimes not enough …

## Isomeric ions



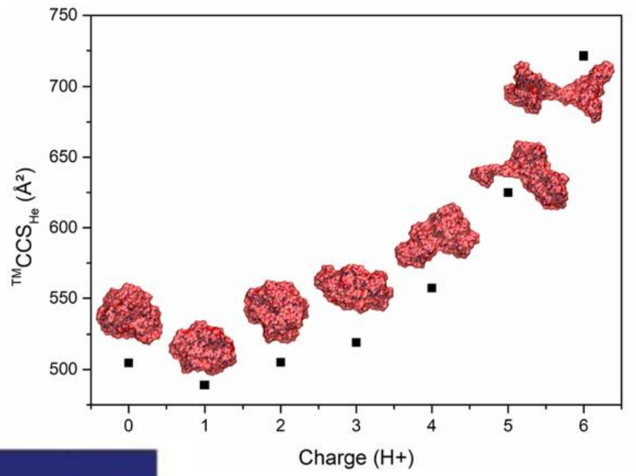
Escin 1b

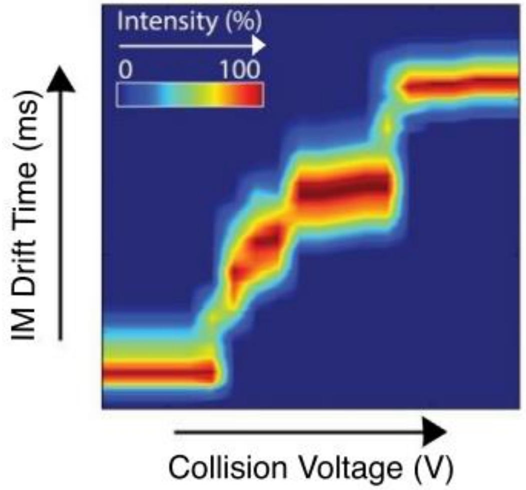


Isoescin 1b

## Shape of ions in gas-phase

« PAMAM » dendrimer

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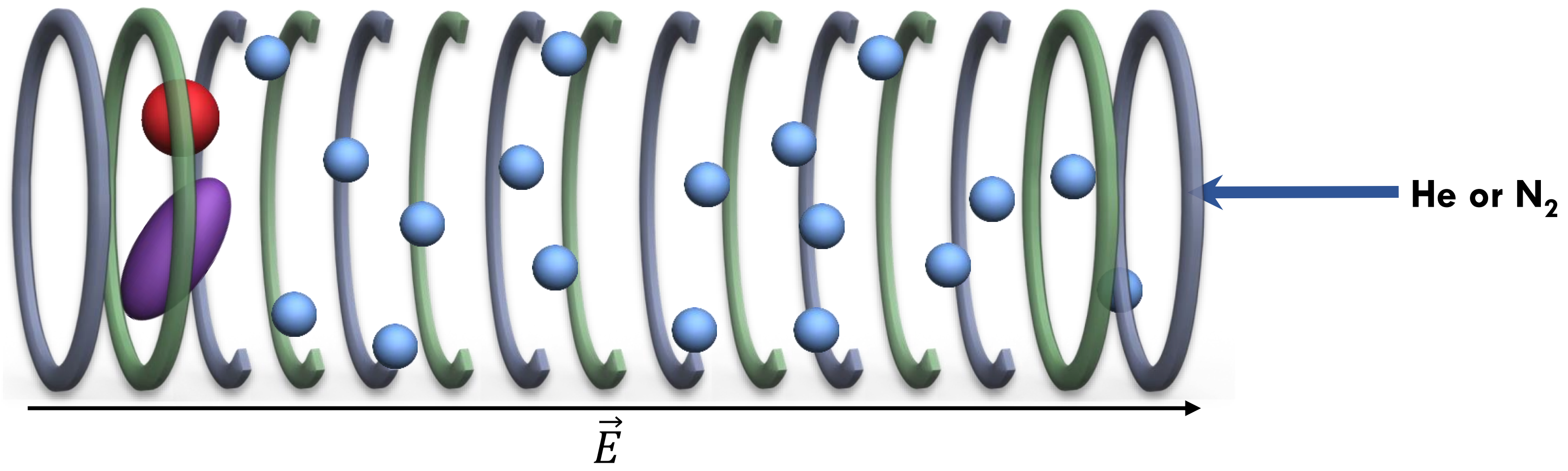
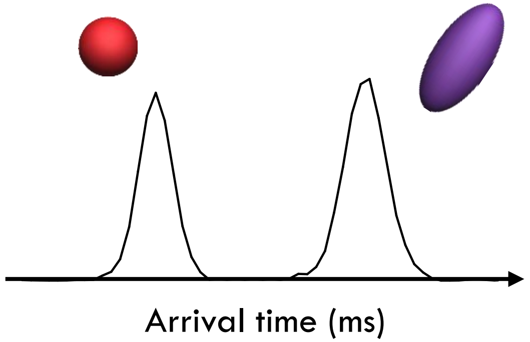
# Ion mobility spectrometry

- Measurement of ion mobilities **K (cm$^2$ V$^{-1}$ s$^{-1}$)** : a physical property of ions in the gas-phase

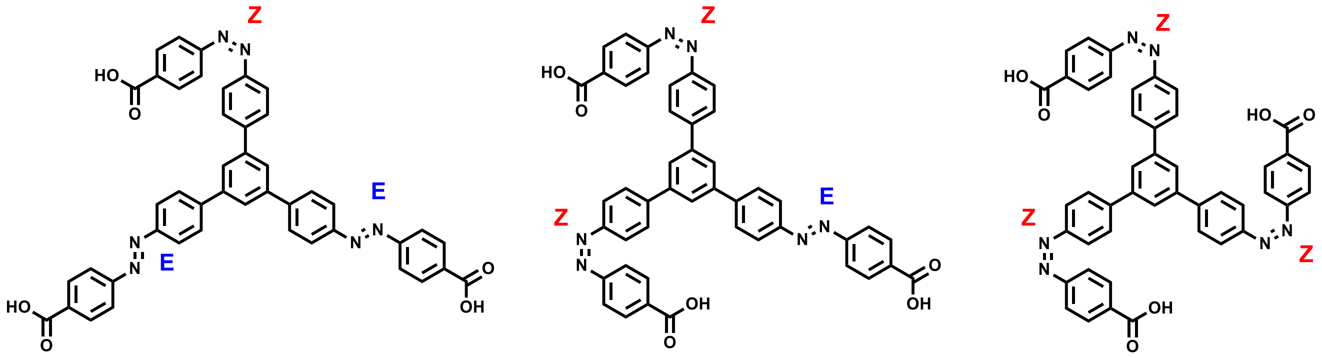



$$v = \frac{L}{t_D} = K.E$$

~ **Motion of ions in the gas**

- Coupling of IMS and MS to cope with our isomer problem !




$$K = \frac{3}{16}\sqrt{\frac{2\pi}{\mu k_B T}}\frac{ze}{N\Omega}$$

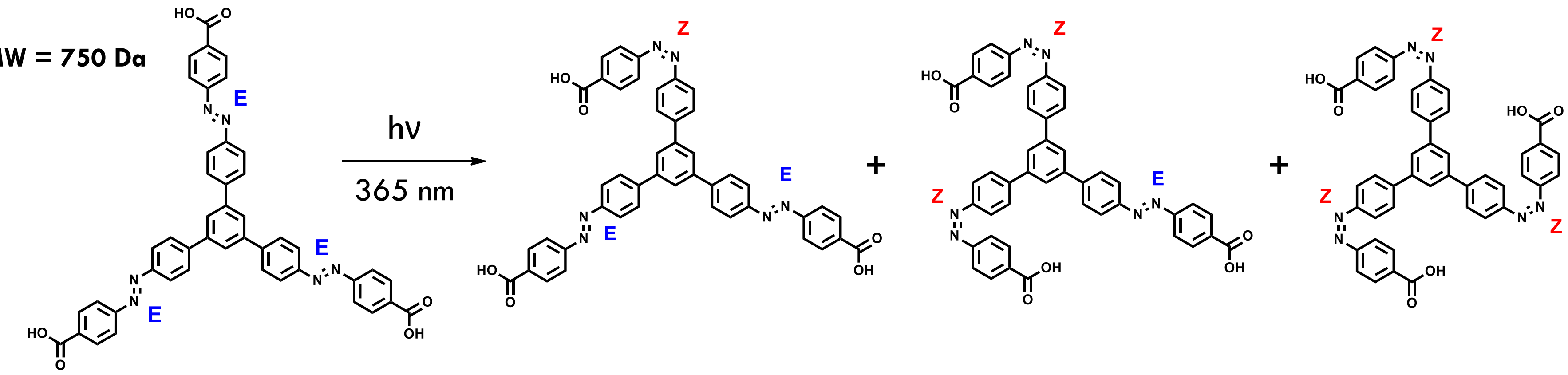
**Ω = Collision cross section (Å²)**

Larriba-Andaluz et al. (2014) *J. Chem. Phys.*, 141 (19), 194107.
D'Atri V. et al. (2015). *J. Mass Spectrom.*, 50 (3), 711-726.

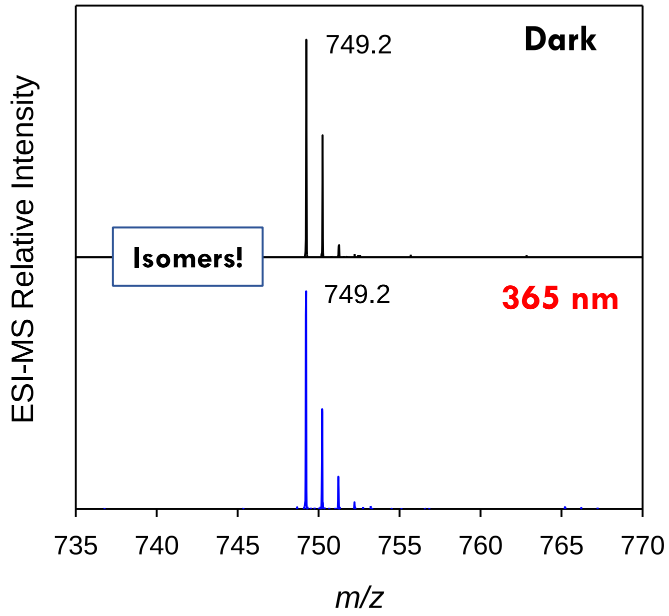
# Ion mobility spectrometry

➢ **Mass spectrometry** : Separation according to $m/z$ ratios // **Ion Mobility Spectrometry** : Separation according to charge, size and shape

MW = 750 Da

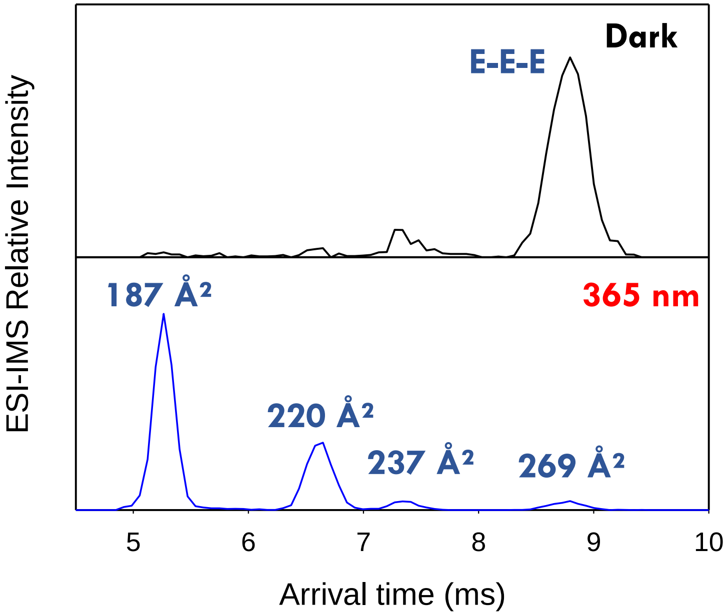



**Electrospray Ionization**
Deprotonation : $[M-H]^-$




**IMS signal**

*m/z* **749.2**

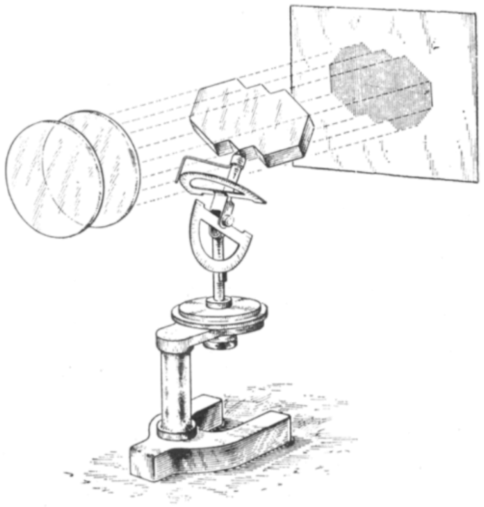




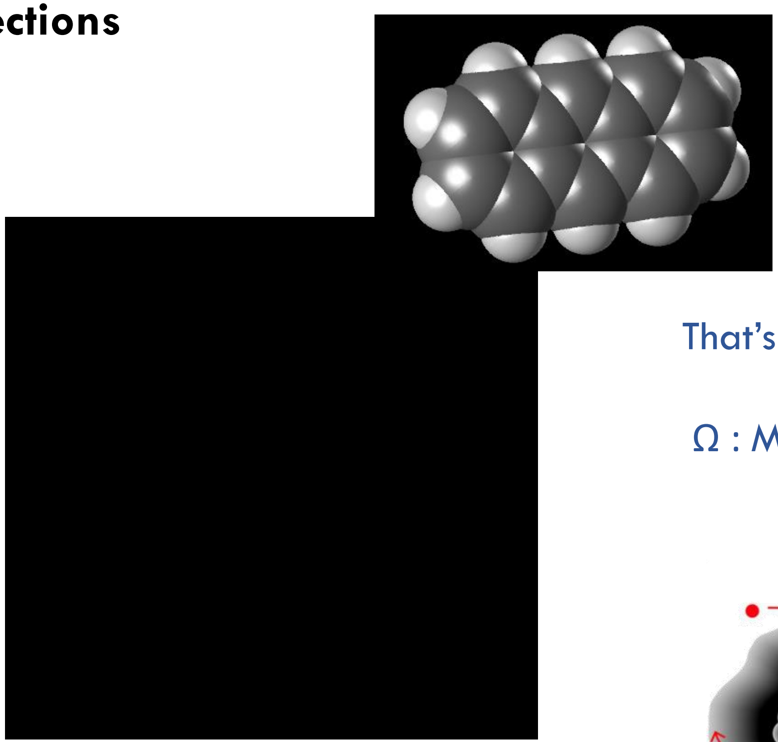

Galanti *et al.* (2018) *J. Am. Chem. Soc.*, 140 (47), 16062-16070.

# Collision cross sections

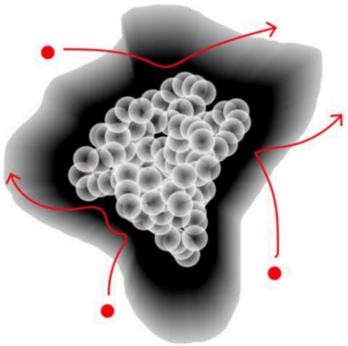
- Bringing a 3D ion shape into a 2D variable (Å²)



**Collision cross section :**
**Orientationally-averaged**
**shadow projection related to ion size and shape**



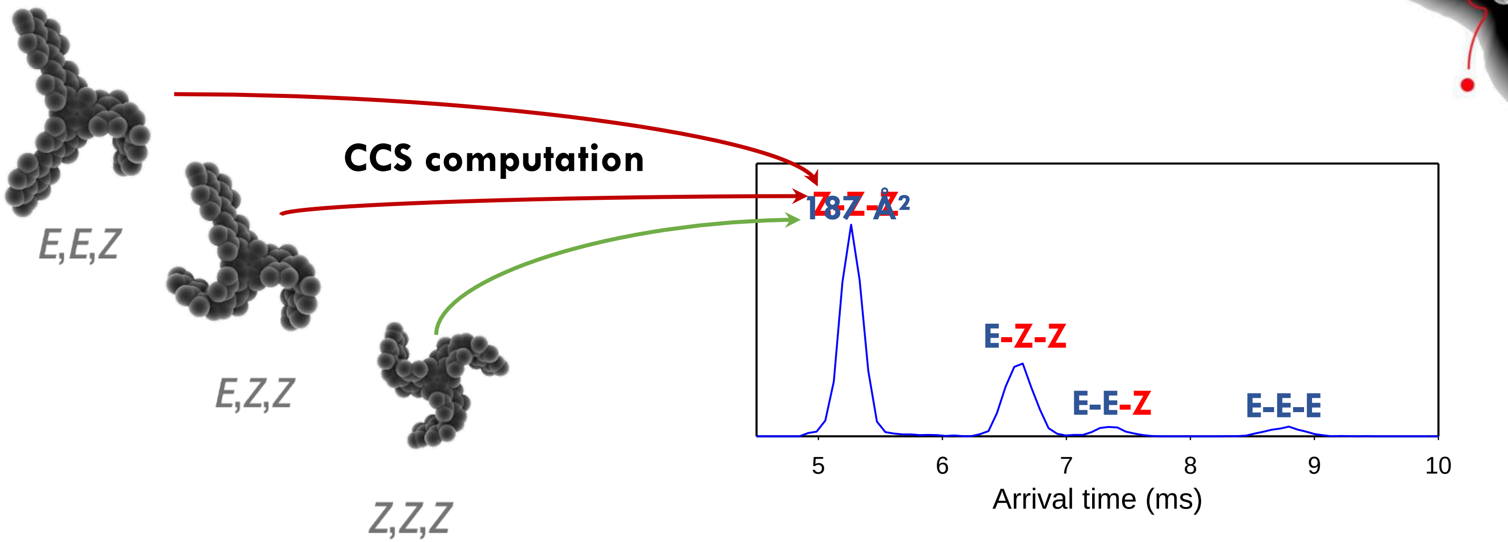
That's the easy way ! ;-)
Formally…
Ω : Momentum transfer cross-section



- 1) CCS for comparison with theory

**Molecular dynamics**

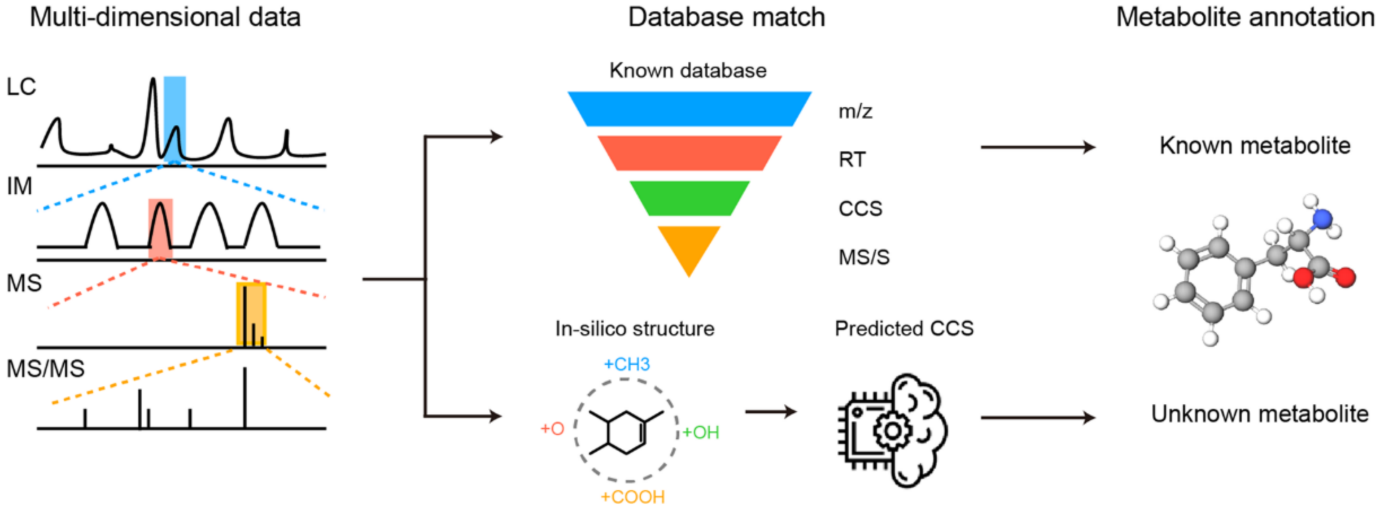
**DFT**



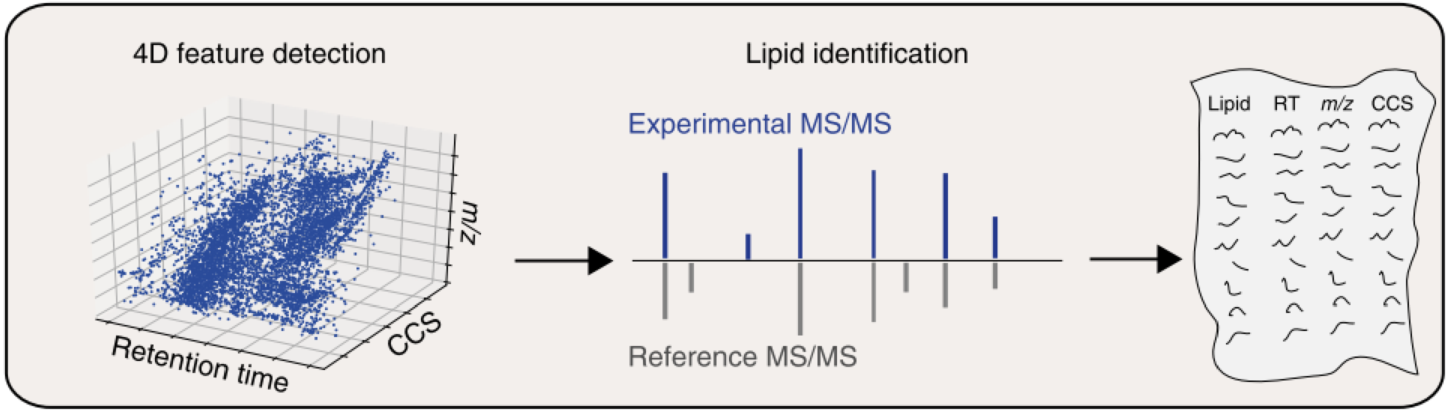

CCS simulations made with IMOs (https://www.imospedia.com/)
Mack, E. et al. (1925) *J. Am. Chem. Soc.*, 47(10), 2468-2482.
Larriba-Andaluz et al. (2014) *J. Chem. Phys.*, 141 (19), 194107.
D'Atri V. et al. (2015). *J. Mass Spectrom.*, 50 (3), 711-726.

# Collision cross sections

- **2)** CCS for "omics" - small molecule identification using libraries




Data analysis and lipid identification



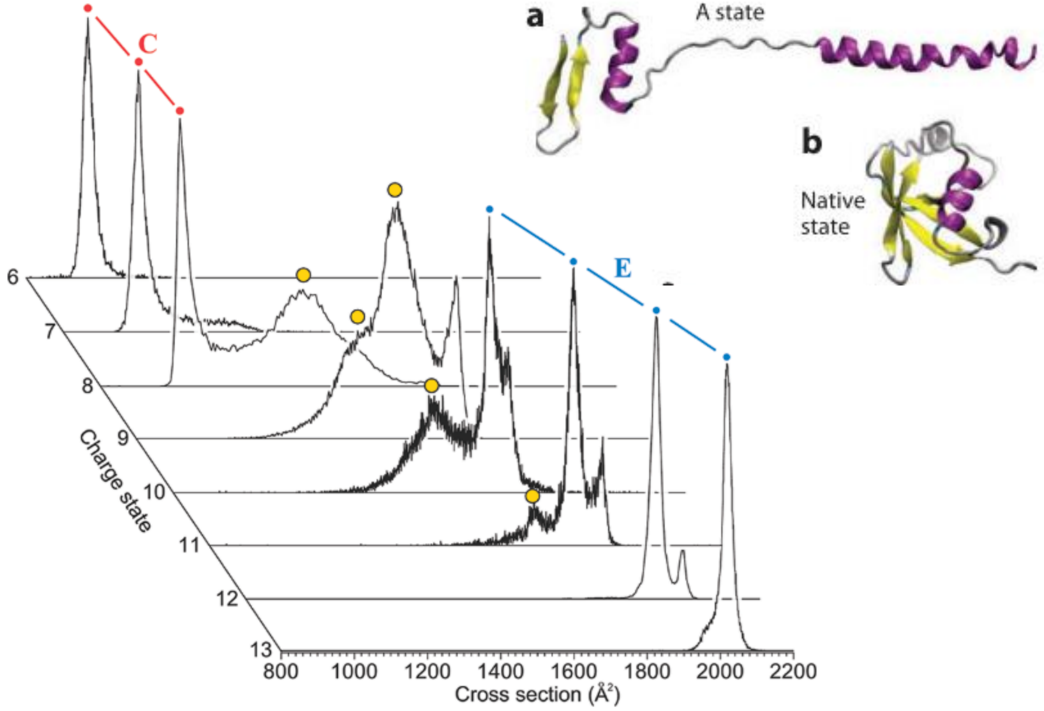


Luo, M et al. (2020) *J. Anal. Test.*, 4 (3), 163-174.
Vasilopoulou C. et al. (2020). *Nat Commun.*, 11 (1), 331.

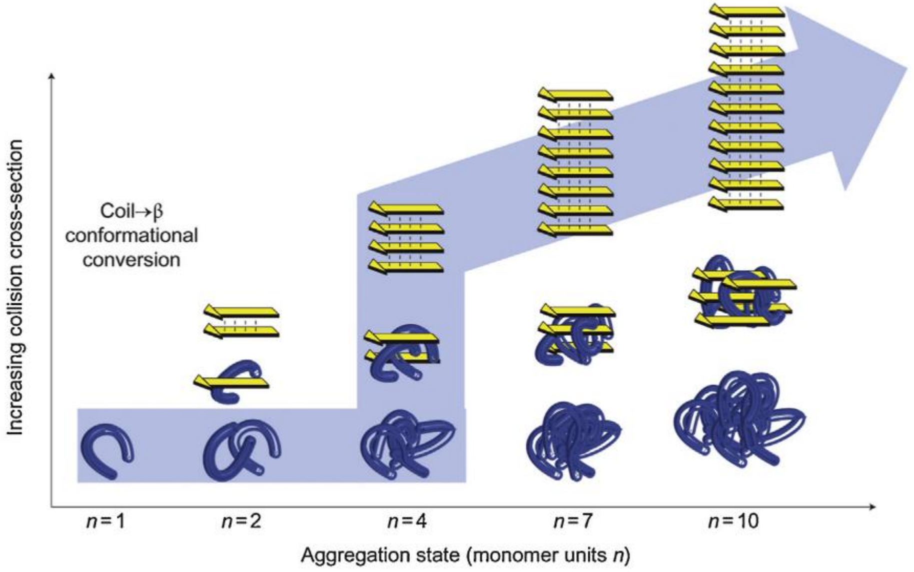
# Collision cross sections

- **3)** CCS to unravel structural features

**Ubiquitin**




**Peptide aggregation**





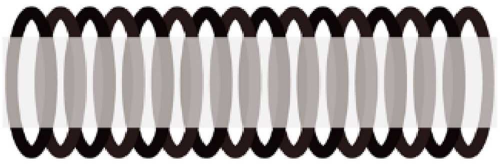
Wyttenbach, T and Bowers, M. (2011) *J. Phys. Chem B..*, 115 (42), 12266-12275.
Bleiholder, C. et al. (2003). *Nat Chem.*, 3, 172-177.

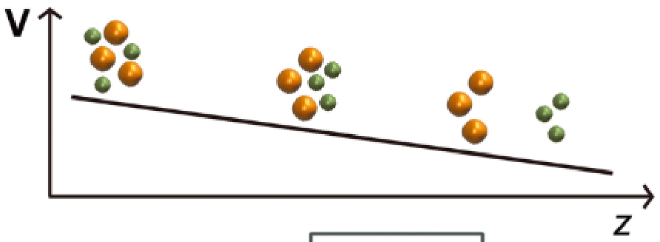
# Outlook

- Why would you do **ion mobility** ?
- Ion mobility : **Principle** of the experiment & **collision cross sections**
- Instrumentation : **DTIMS, TWIMS, TIMS**

# Instrumentation

- « Historic » design : **Drift tube (DTIMS)**





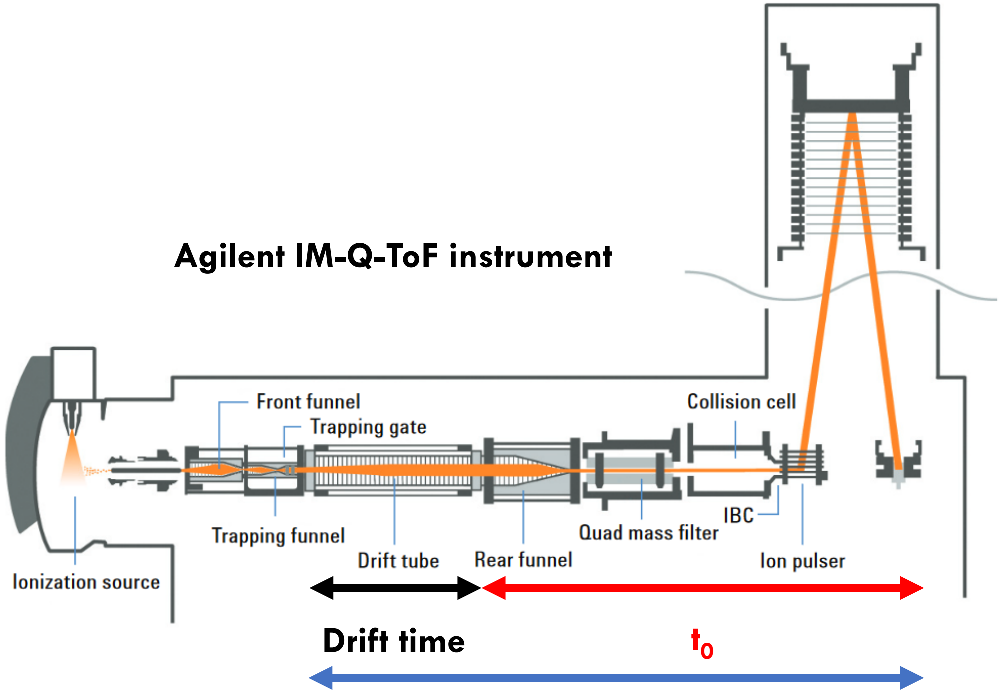

Drift time

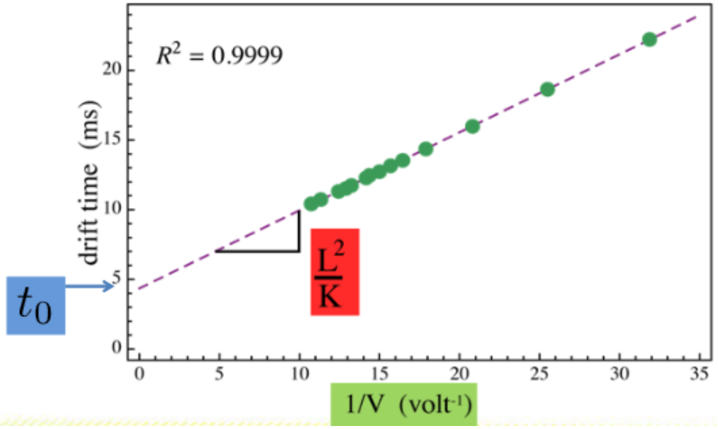
Arrival time

$$v = \frac{L}{t_D} = K.E \longrightarrow t_A = tD + t_0$$

$$t_A = \frac{L}{KE} + t_0 = \frac{L^2}{KV} + t_0$$

**Agilent IM-Q-ToF instrument**

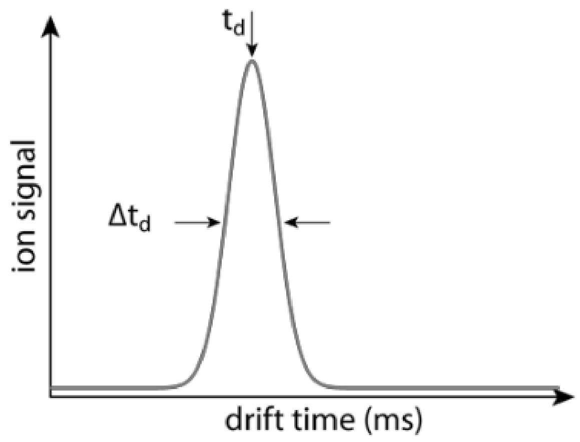



D'Atri V. et al. (2015). *J. Mass Spectrom.*, 50 (3), 711-726.
https://www.agilent.com/cs/library/technicaloverviews/public/5991-3244EN.pdf

# Instrumentation

- « Historic » design : Drift tube (DTIMS) – Resolution ??




$$R_{DT} = \frac{t_d}{\Delta t} = \frac{1}{4}\sqrt{\frac{q}{k_B \ln 2}}\sqrt{\frac{V_d}{T}}$$

$V = E.L$

Resolution mainly limited by diffusion in the gas !

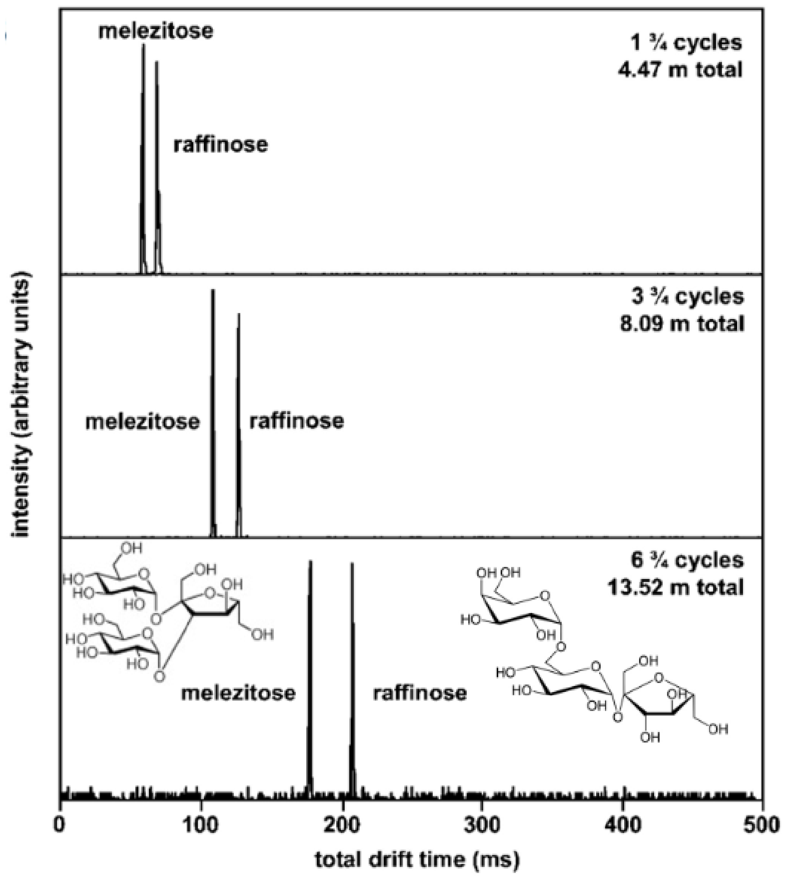
**Increasing R ??**

**Increasing L** | **Increasing E** | **Decreasing T**

Merenboom, S. et al. (2009). *Anal. Chem.*, 81, 1482-1487.

# Instrumentation

➤ « Historic » design : Drift tube (DTIMS) – Resolution ??




$$R_{\mathrm{DT}} = \frac{t_{\mathrm{d}}}{\Delta t} = \frac{1}{4}\sqrt{\frac{q}{k_{\mathrm{B}}\ln 2}}\sqrt{\frac{V_{\mathrm{d}}}{T}}$$

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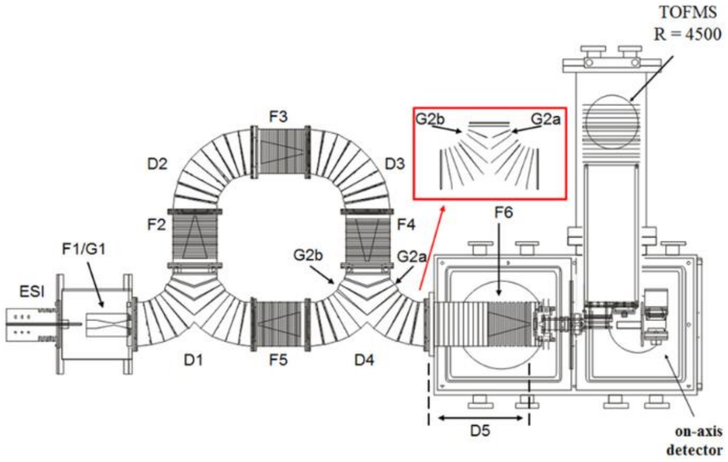
Resolution mainly limited by diffusion in the gas !

**Increasing R ??**

Increasing L | Increasing E | Decreasing T




Merenboom, S. et al. (2009). *Anal. Chem.*, 81, 1482-1487.

# Instrumentation

- « Historic » design : Drift tube (DTIMS) – Resolution ??

- Increasing E … **Problem of the « low-field » limit !**

$$v = \frac{L}{t_D} = K.E$$

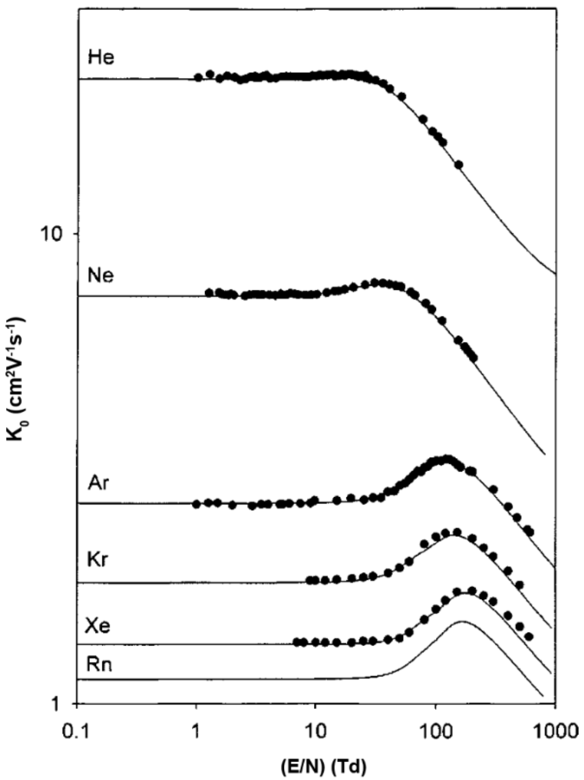
$$K = \frac{3}{16}\sqrt{\frac{2\pi}{\mu k_B T}}\frac{ze}{N\Omega}$$

**Exact provided that thermal energy >> energy gained from the electric field**

**Key parameter : E/N**

**(Think about difference between IMS and CID !)**

Mobility of $K^+$ in various gases




$$R_{DT} = \frac{t_d}{\Delta t} = \frac{1}{4}\sqrt{\frac{q}{k_B \ln 2}}\sqrt{\frac{V_d}{T}}$$

$$V = E.L$$

Resolution mainly limited by diffusion in the gas !

**Increasing R ??**

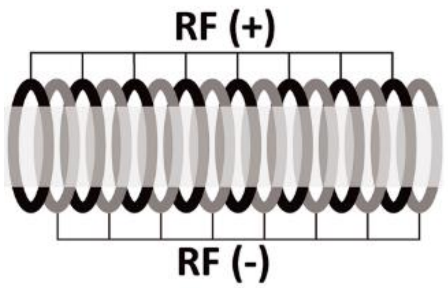
Increasing L | Increasing E | Decreasing T

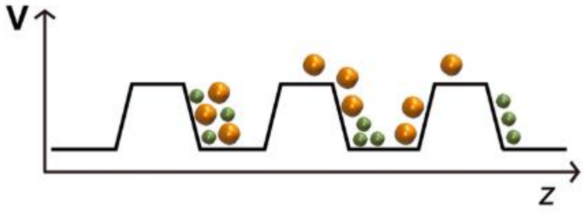


Viehland, L. et al. (2005). *J. Phys. Chem.*, 121, 341-351.

# Instrumentation

➢ First commercial IMS : Waters Synapt – **Traveling wave (TWIMS)**



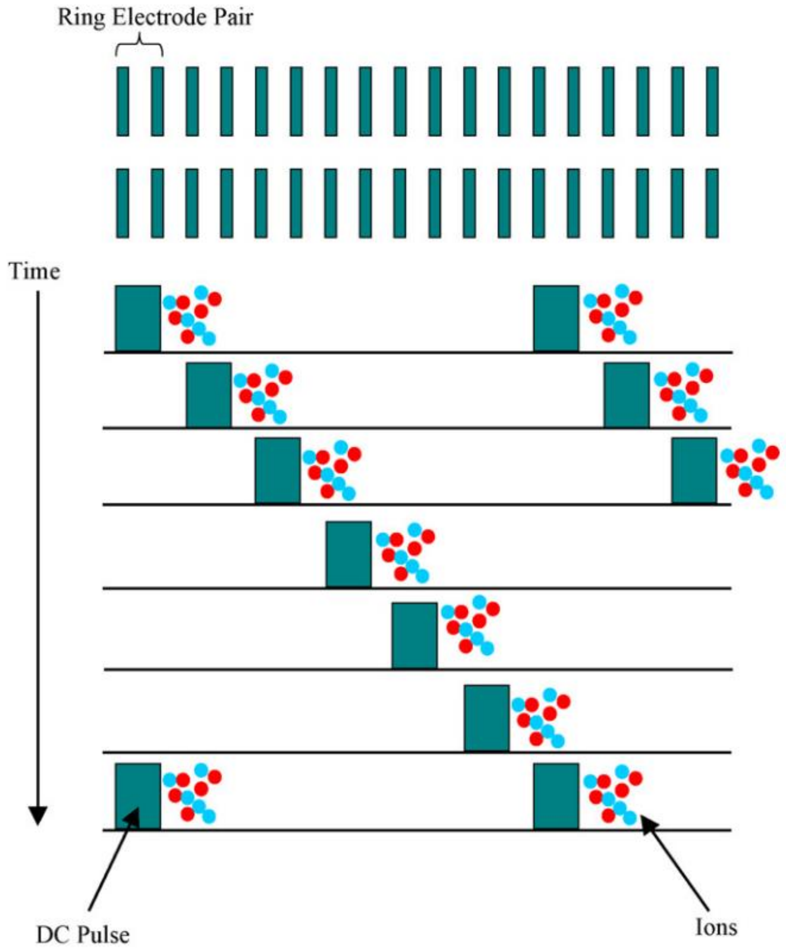

**RF potential (to confine) + moving DC pulse (to push) = Traveling wave**

In TWIMS :

$CCS \sim tD$ → $CCS \sim tDA$

**E not constant !**
**Not possible to apply Mason-Schamp equation !**

An external calibration is required to determine CCS from TWIMS measurements

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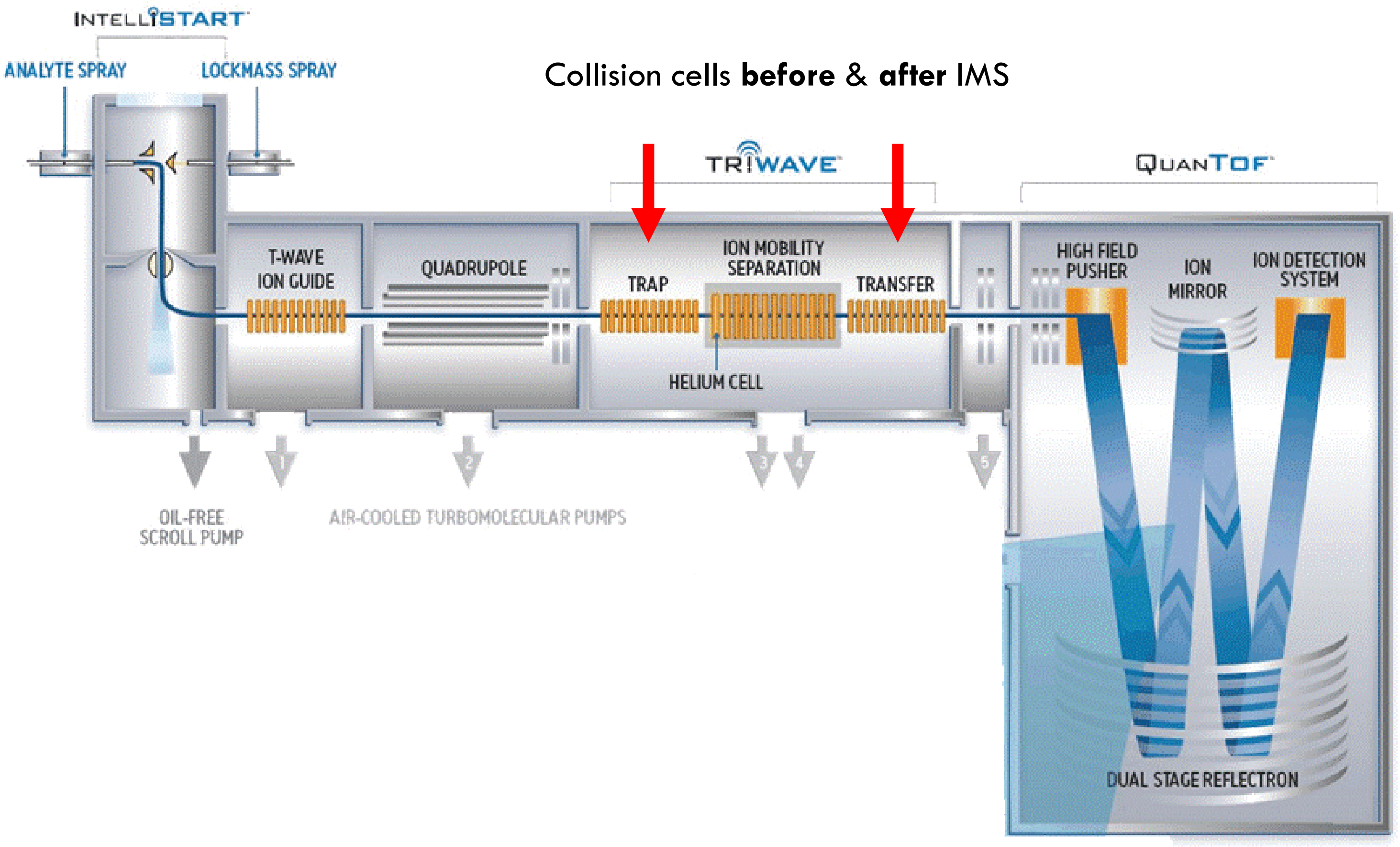
Giles, K. et al. (2007) *Int. J. Mass. Spectrom.*, 261 (1), 1-12.

# Instrumentation

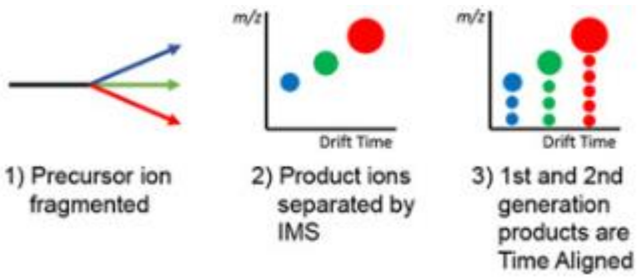
- First commercial IMS : Waters Synapt – **Traveling wave (TWIMS)**

Collision cells **before** & **after** IMS




Possible to do "MS³" provided that first daughter ions have different mobilities !!

"Time aligned parallel fragmentation"





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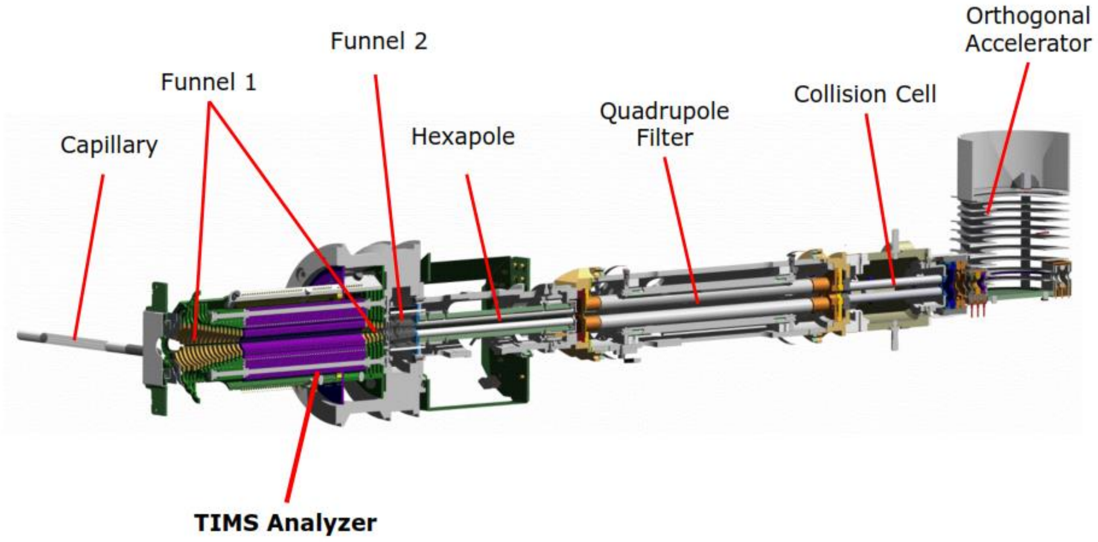
Giles, K. et al. (2007) *Int. J. Mass. Spectrom.*, 261 (1), 1-12.

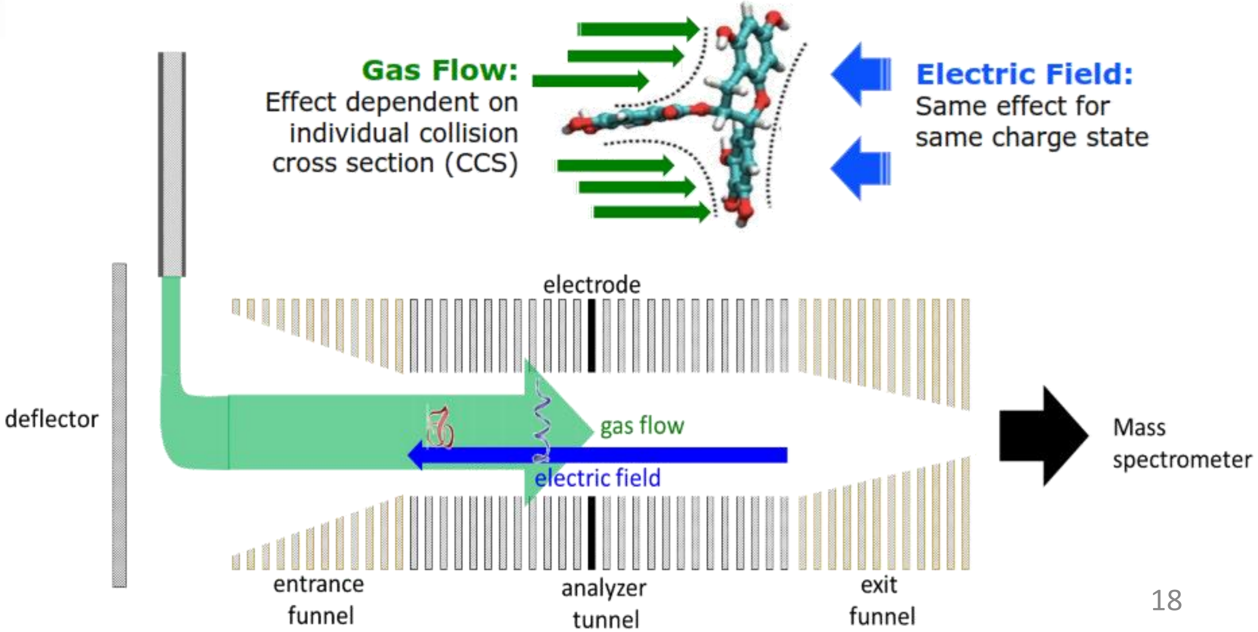
# Instrumentation

- Another commercial IMS : Bruker timsToF – **Trapped IMS (TIMS)**





**Accumulate, trap, and elute principle to enhance sensitivity & resolution**

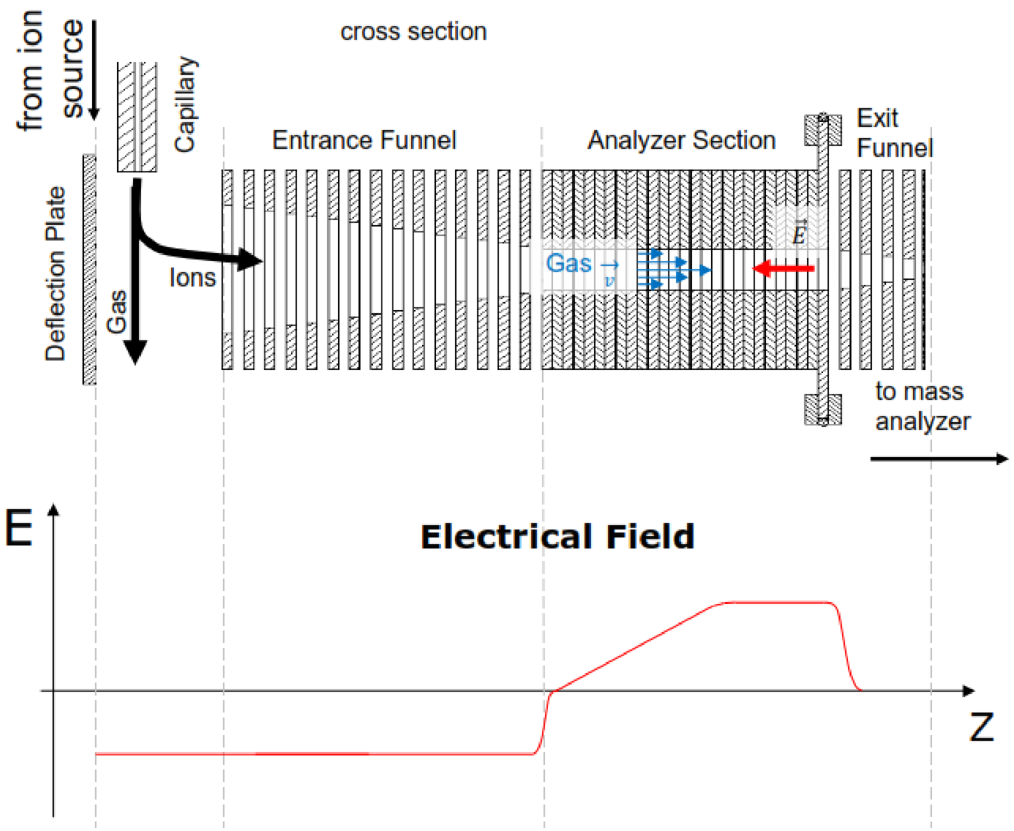


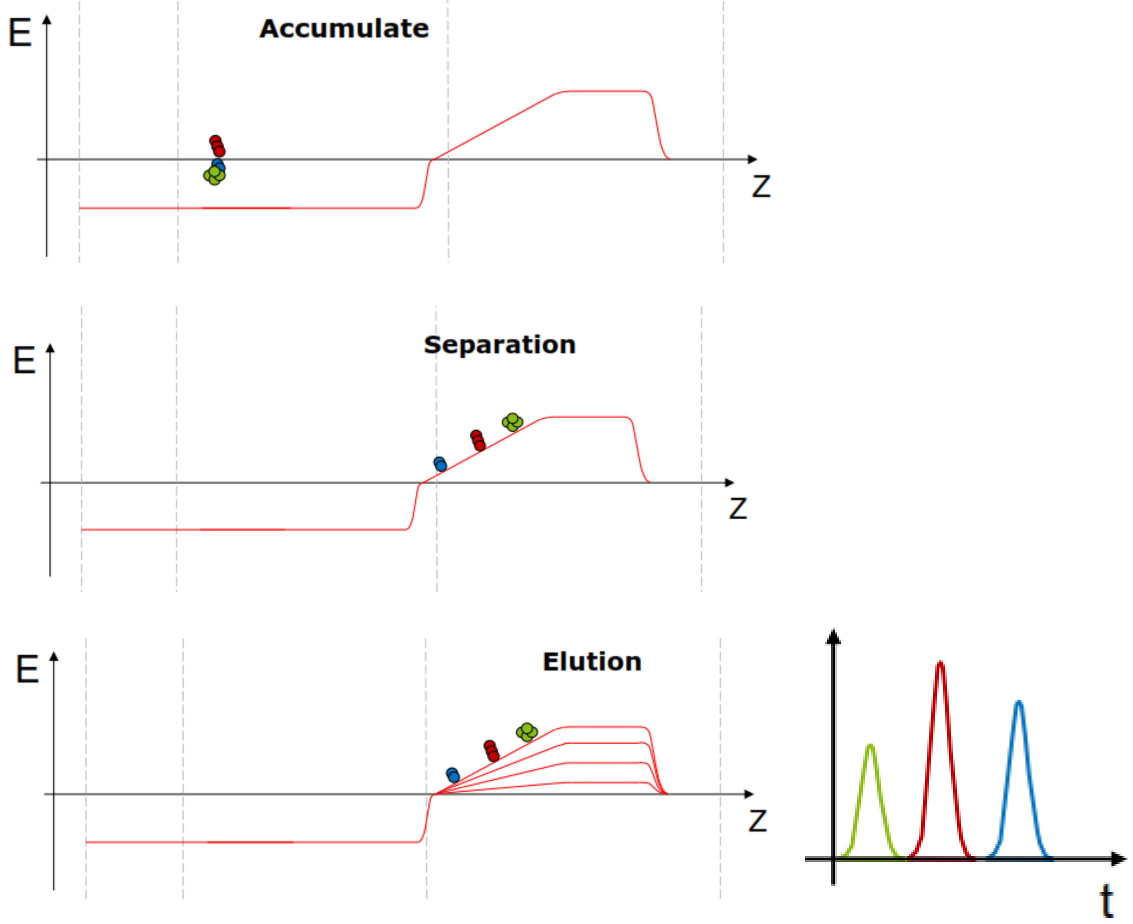



Bruker training material

# Instrumentation

- Another commercial IMS : Bruker timsToF – **Trapped IMS (TIMS)**




**Think about which ion elutes first !**

# Instrumentation

- Comparison between the three main setups

**Estimates of typical operating parameters for contemporary commercial instruments**

| Instrument and manufacturer | Operation principle | $p$ (Torr) | $E$ (V/cm) | Typical $E/N$ (Td) | Typical $v_D$ (m/s) |
|---|---|---|---|---|---|
| 6560 IMS-Q-TOF, Agilent Technologies | Drift tube | 4 | 9.5–20 | 7–15 [33] | 10–80 |
| Synapt HDMS, Waters | TWIMS | 0.4 | 21 (maximum axial field at wave height = 10 V) [34] | ≤160 | 200–600 [29] |
| Synapt G2, G2-S and G2Si HDMS, Waters | TWIMS | 2 | 100 (maximum axial field at wave height = 40 V) | ≤155 | 200–300 [30] |
| TIMS-TOF, Bruker | TIMS | 2 | 30–55 | 45–85 | 120–170[a] [35] |
| IMS-TOF, Tofwerk | Drift tube | 570–788 | ~400 | 1–2 [36] | ~5 |

[a] Gas velocity; in TIMS the ion is static.

**Resolution**

**~ 80**

**~ 40**

**~ 200**